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混合粉末を、その混合粉末の組成比に
置換させ、その置換した混合粉末を製造した。
実施例 2

UGLA、AALA、及びBALAをそれぞれ異なる
重量の割合で混合した。その混合粉末は、その混合
粉末の組成比に置換させ、その置換した混合粉末を
製造した。その置換した混合粉末を製造した。
その置換した混合粉末を製造した。

実施例 3

このA、UGLA、AALA、及びBALAのそれぞれ
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実施例 4

カルボニルニラ、クロロカタとAALG (PBR
48703) の各異性体より得られた固体化合物を
その置換した混合粉末の組成比に置換させ、
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実施例 5

このAニラ、BALAニラ、UGLAニラ、
AALAニラ、BPAニラをそれぞれ異なる
重量の割合で混合した。その置換した混合粉末
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混合粉末を製造した。その置換した混合粉末を
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実施例 6

カルボニルニラ、クロロカタとAALG (PBR
48703) の各異性体より得られた固体化合物を
その置換した混合粉末の組成比に置換させ、
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54-TITLE OF THE INVENTION

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English Title: MANUFACTURED MILK TO WHICH A HIGH-LEVEL
UNSATURATED FATTY ACID COMPONENT HAS BEEN
ADDED

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Specification

1. Title of Invention:

Highly unsaturated fatty acid components added to synthetic milk

2. Scope of Patent Claim(s)

1. This is a method for the manufacture of highly unsaturated fatty acid components added to synthetic milk wherein it is composed of (eicosadienic acid), (bishomo- γ -linolenic acid) arachidonic acid or (ecosapentanic acid), or an ester of any of the above, their fats or oils which contain the fatty acid, a hydrolysate of any of the above, or esterfied materials of said fat-splitting products, either mixed by themselves or in combination with the others. In addition, the γ -linolenic acid, an ester of said fatty acid, the fats and oils which contain said fatty acid, the hydrolysate of said fat and oil, or the esterfied materials of said fat-splitting products are added to the above mixture.

3. Description of invention

Industrial applications

This invention is concerned with a method for manufacturing synthetic milk in which the powdered milk or liquid milk within the synthetic milk as well as minute fatty acid components are strengthened.

Prior art technology

The γ - linolenic acid, (ecosadienic acid), (bishomo- γ - linolenic acid, arachidonic acid, and eicosapentanoic acid (abbreviated hereafter as GLA, EDA, OGLA, ARA, and EPA) are fatty acids which are indispensable to highly developed animals. In living organisms they are the bodies of the (prostaglandin) group, performing such important functions as the regulation of blood pressure, and hormone secretion control. They also represent the highly unsaturated fatty acids which possess physiological activities, and are induced from the linoleic acid and the γ - linolenic acid which constitute the indispensable fatty acids by means of the Δ^6 or Δ^5 desaturase and a carbonic elongation enzyme. It has been known that, among the above, the activity of desaturase is weakened by senility cancer, diabetes, and other diseases. As a result, the production of (prostaglandin) is suppressed, resulting in a number of health

problems. Therefore, it is inappropriate to take this highly unsaturated fatty acid directly for remedial purposes or as a preventative measure.

In new-born infants these highly unsaturated fatty acids are received from the mother's milk. The various (prostaglandin) which is induced from highly unsaturated fatty acids are thought to contribute to immunity as a final function. With regard to the newborn, these components are absorbed from the mother and are exceedingly important from the standpoint of maintaining life.

However, the amount of the various fatty acids contained in natural human milk is not accurately known. Moreover, since, among other things, said various fatty acids have exceptionally high values, in such man made milk as powdered milk or liquid milk, although minute quantities of various fatty acids can be added, it has been difficult to assure that the fatty acids in powdered milk or liquid milk is equal to or in excess of that found in the milk of nursing mothers.

Problems overcome by this invention

For the above reasons, the purpose of this invention is to provide milk which contains a fatty acid structure which is

similar to that of human milk. The synthetic milk described by this invention contains minute quantities of said fatty acids.

Methods for solving said problems

The inventors have attempted to try to clarify the amino acid structure in the human milk and the fatty acid structure in powdered milk produced by ordinary means. By comparing the two, it was understood that GLA, EDA, CGLA, ARA and EPA are lacking in powdered milk. Furthermore, a method was sought to produce these fatty acids less expensively, using the fermentation method, thus completing the invention.

Therefore, this invention provides synthetic milk wherein it is composed of (eicosadienic acid), (bishomo- γ -linolenic acid) arachidonic acid or (ecosapentanic acid), or an ester of any of the above, their fats or oils which contain the fatty acid, a hydrolysate of any of the above, or esterfied materials of said fat-splitting products, either mixed by themselves or in combination with the others. In addition, the γ -linolenic acid, an ester of said fatty acid, the fats and oils which contain said fatty acid, the hydrolysate of said fat and oil, or the esterfied materials of said fat-splitting products are added to the above mixture.

Explanation of actual procedure

First of all the fatty acid structure of human milk (3 months after the birth of the baby) and two types of commercially available powdered milk (made into liquid form with a concentration of 13 g/100 ml), is shown in the following chart.

Fatty Acid	Human Milk	Powdered Milk	
	(mg / ml)	A (mg/ml)	B
Myristic acid	2.0	0.9	2.3
Palmitic acid	5.4	6.2	5.7
(Palmitoleic acid)	1.0	0.8	0.2
Stearic acid	2.1	2.8	1.3
Oleic acid	9.4	10.2	7.5
Linoleic acid	4.4	5.8	5.3
α - linolenic acid	0.9	0.9	0.5
τ - linolenic acid	0.03	tr	tr
(Eicosadienic acid)	0.08	0	0
(Bishomo) - τ - linolenic acid	0.08	0	0
Arachidonic acid	0.3	tr	tr
(Eicosapentanic) acid	tr	tr	0
(Docosahexanic) acid	tr	0.1	0

(Note: some substances are phonetic)

When the mother's milk and the synthetic milk are compared, the minute quantities of fatty acid such as τ - linolenic acid,

(eicosadienic acid), (bishomo) 231 - linolenic acid, arachidonic acid and (eicosapentanic) acid are lacking in the synthetic milk. For that reason, with this invention, the fatty acids discussed above are added during the manufacturing process of the synthetic milk, or in the finished product, thereby obtaining a synthetic milk which possesses a structure containing minute quantities of fatty acid.

In this case, the quantity of added fatty acid differs depending upon various conditions. For example, it is thought that the fatty acid structure in the human milk changes by the time it is passed after the birth of the baby. Therefore, the added quantity of the fatty acid differs depending upon the age of the baby. In addition,, the material and the manufacturing process of the synthetic milk also differs. Hence, in the manufacturing process described by this invention the fatty acid discussed above or the materials containing the fatty acids are added by themselves into the mixture, depending upon the conditions.

For example, ϵ - linolenic acid at 0.02-0.03%; (eicosadiennic accid) at 0.05-0.08%, (bishomo) - ϵ - linolenic acid at 0.05-0.08%; arachidonic acid at 0.2-0.3%; and (eicosapentanic acid) at 0.01-0.03% are added to the dry product.

SPECIFICATIONS

1. Title of the Invention

Manufactured Milk To Which a High-Level Unsaturated Fatty Acid Component Has Been Added

2. Claim

1. Manufactured milk in which there has been added, either alone or in a combination, eicosadienoic acid, Bis-homo- γ -linoleic acid, arachidonic acid, or eicosapentaenoic acid, esters of the aforesaid fatty acids, oils and fats contained in the aforesaid fatty acids, or hydrolysates of the aforesaid fats and oils or an esterified product of the dissolved matter of the aforesaid fats and oils, or, in which there has been added to those materials γ -linoleic acid, esters of the fatty acids, oils and fats containing the fatty acids, or hydrolysates of the fatty acids or an esterified product of the dissolved matter of the fats and oils.

3. Detailed Specifications

The invention under review pertains to manufactured milk in which a minute amount of a fatty acid component, which is lacking or insufficient in manufactured milk, such as powdered milk or liquid milk, has been reinforced.

(Traditional Technology)

γ -linoleic acid, eicosadienoic acid, Bis-homo- γ -linoleic acid, arachidonic acid, and eicosapentaenoic acid (hereinafter these fatty acids are occasionally referred to as "GLA, EDA,

DGLA, ARA, and EPA") are indispensable fatty acids in sophisticated animals. In human beings, they are the starting materials in the creation of prostaglandins, which perform important functions, such as regulation of blood pressure and regulation of hormone secretion; prostaglandins are themselves high-level unsaturated fatty acids that are physiologically active. Prostaglandins are derived from linoleic acid or α -linoleic acid, which are essential fatty acids, by Δ^6 -desaturase or Δ^5 -desaturase and a carbon-chained elongation enzyme. The activity of the desaturases can be weakened because of aging, cancer, diabetes, and other illnesses and phenomena, and, as a result, the production of prostaglandins may be hindered. It is commonly known that if the production of prostaglandins is thwarted, that various risks to health can result. Therefore, direct intake of the aforesaid high-level unsaturated fatty acids is useful in the treatment, or in the prevention, of these health risks.

Infants obtain these high-level unsaturated fatty acids from their mothers' milk. Prostaglandins, which are derived from high-level unsaturated fatty acids, also seem to be related to a human body capability to show immunity to certain illnesses. Consequently, intake of the components for prostaglandins from their mothers' milk is certainly crucial to ensuring that newborns will enjoy healthy lives.

Nevertheless, researchers do not know for sure how much of the aforesaid fatty acids is contained in natural mothers' milk.

In addition, in that the aforesaid fatty acids are very expensive and there are other factors to consider, it is difficult to add a minute amount of fatty acids to manufactured milk, such as powdered milk or liquid milk, and it is difficult to produce powdered milk or liquid milk that contains the same quantity, or a higher quantity, of the fatty acids that are found in mothers' milk.

(Problem Points that the Invention Will Solve)

As a result of the invention under review, the minute quantity of fatty acids is made stronger, and milk that has a fatty acid component that is approximately the same as that found in human, natural mothers' milk is obtained.

(Procedures to Solve the Problem Points)

The inventors did studies on the amino acid component in human, natural milk and the fatty acid component in milk produced by conventional methods. When comparison was made, they determined whether the GLA, the DGLA, the ARA, or the EPA was sufficient in powdered milk. In addition, through another invention, the inventors had invented a method of producing these fatty acids by a method of fermentation at a low cost. As a result of these efforts, the invention under review was perfected.

Therefore, the invention under review provides manufactured milk, such as powdered milk or liquid milk, in which there has been added, either alone or in a combination, eicosadienoic acid, Bis-homo- γ -linoleic acid, arachidonic acid, or eicosapentaenoic

acid, esters of the aforesaid fatty acids, oils and fats contained in the aforesaid fatty acids, or hydrolysates of the aforesaid fats and oils or an esterified product of dissolved matter of the aforesaid fats and oils, or, in which there has been added to those materials γ -linoleic acid, esters of the fatty acids, oils and fats containing the fatty acids, or hydrolysates of the fatty acids or an esterified product of dissolved matter of the fats and oils to those materials.

(A Detailed Explanation)

The following table shows the fatty acid composition in human mothers' milk (five months after childbirth) and two types of milk (ones where a concentration of 13 g/100 ml was dissolved in water) that are available on the market.

Fatty Acids	Mothers' Milk (mg/ml)	Powdered Milk	
		A	B
		(mg/ml)	
Myristic acid	2.0	0.9	2.3
Palmitic acid	5.4	6.2	5.7
Palmitoleic acid	1.0	0.8	0.2
Stearic acid	2.1	2.8	1.3
Oleic acid	9.4	10.2	7.5
Linoleic acid	1.4	5.8	5.3
α -linoleic acid	0.9	0.9	0.5
γ -linoleic acid	0.03	tr	tr
Eicosadienoic acid	0.08	0	0

Bis-homo- γ -linoleic acid	0.08	0	0
Arachidonic acid	0.3	tr	tr
Eicosadienoic acid	tr	tr	0
Docosahexenoic acid	tr	0.1	0

When comparison is made between the natural mothers' milk and the manufactured milk, it is found that the fatty acids, which are in a minute amount, such as linoleic acid, eicosadienoic acid, Bis-homo- γ -linoleic acid, arachidonic acid, and eicosapentaenoic acid, are not sufficient in the manufactured milk. Therefore, in the invention under review, fatty acids, like those indicated above, are added to the granules in the processes of making the milk or to the finished product. As a result, manufactured milk that has a fatty acid content that is approximately the same as the fatty acid content in mothers' milk will be obtained.

The amount of the aforesaid fatty acids to be added will depend upon various conditions. For example, the fatty acid composition in human mothers' milk seems to change as the time since childbirth grows longer. Therefore, the amount of fatty acids to be added will depend upon when after birth the manufactured milk is to be administered to the infant. The ingredients of the manufactured milk will also depend upon the production processes employed in the production of the manufactured milk. Therefore, in the invention under review, the aforesaid fatty acids or matter containing the fatty acids may be

added by themselves or in combinations in accordance with the conditions at hand.

For example, to a dry product may be added 0.02-0.03% γ -linoleic acid, 0.05-0.09% eicosadienoic acid, 0.05-0.08% Bis-homo- γ -linoleic acid, 0.2-0.3% arachidonic acid, or 0.01-0.03% eicosapentaenoic acid. In addition, the proper combination of matter containing complex fatty acids, for example, the utilization of lipids or hydrolysates of lipids, and a single fatty acid will strengthen the fatty acid content to the desired level. The quantity of the fatty acid or the ester of a fatty acid should be a 0.001-2 weight % for powdered milk, but it is recommended that that quantity be 0.0001-0.2% for liquid milk.

The aforesaid fatty acid can be added in a number of forms. For example, it can be added in granulated or dissolved form, or it can be added as a salt of the fatty acid, such as a sodium salt or a potassium salt. The fatty acid can also be added as an ester, such as a methyl ester or an ethyl ester. In addition, lipids that contain the aforesaid fatty acids in a high ratio, for example, triglyceride or a hydrolysate of triglyceride, or esterified products of a hydrolysate, such as esterified methyl or esterified ethyl, are examples of forms that can be utilized.

When the aforesaid fatty acids are used by themselves as individual fatty acids or two or more types of them are combined, products that have been made by acceptable methods can be put to use. For example, additives can be produced by yeast methods or fermentation methods using *Mortierella* microorganisms that have a

high capacity to produce the aforesaid unsaturated fatty acids. For example, after *Mortierella* microorganisms have been cultured and the cultured bacteria has been dried as required, it will be extracted by an organic solvent. As a result, the lipid, which is produced by evaporating, drying, and solidifying the extract, will contain the aforesaid unsaturated fatty acid in a high ratio. This lipid can be utilized as the base material for the fatty acid that pertains to the invention under review. In addition, hydrolysis of this lipid using conventional methods will produce a fatty acid compound or a fatty acid salt compound, such as a sodium salt compound. These types of compounds can then be utilized as the base material for the fatty acid that pertains to the invention under review. The esterification of these fatty acid compounds using conventional methods will produce compounds of a fatty acid ester, e.g., methyl ester or ethyl ester. These substances can then be utilized as the base material for the fatty acid that pertains to the invention under review. Similarly, after isolation of the fatty acid compounds or the fatty acid ester compounds as single fatty acids or fatty acid salts or fatty acid esters, these materials can then be utilized.

The aforesaid fatty acids or the salts of those fatty acids or fatty acid esters or compounds of them can be utilized without further processing or modification. However, so that the substances will have a higher level of consistency, it would be a good idea to add the substance to powdered milk or liquid milk

after those substances have been taken into cyclodextrin. Either an α or a β cyclodextrin can be utilized. From a GLA, EDA, DGLA, an ARA or an EPA fatty acid or from a fatty acid ester, the synthesis of the substance that will be taken into cyclodextrin will be as follows. GLA, EDA, DGLA, ARA, or EPA, in a specified quantity, in the form of a fatty acid or in the form of a fatty acid ester in a saturated or super-saturated aqueous solution of cyclodextrin, will be added. A substance that is taken into cyclodextrin will be produced as a deposit as a result of mixing lasting over a period of ten minutes to ten hours. In the alternative, while a small amount of water is being added to cyclodextrin and the substance is being mixed with a mixer, a specified amount of GLA, EDA, DGLA, ARA, or EPA will be added in the form of a fatty acid or in the form of a fatty acid ester. A substance that is taken into cyclodextrin will be produced as a result of mixing over a period of one to five hours.

If necessity should so dictate, tocopherol sesamol, melanoidins, a flavone derivative, or BHT may be added to the manufactured milk to prevent oxidation. If the milk is to be a powdered milk such additives should be as much as 0.0001-0.1% and if the milk is to be a liquid milk, such additives should be as much as 0.00001-0.01%. The additives mentioned are not the only additives that can be utilized as anti-oxidation agents; any such additives that are commonly known in the industry can also be used.

The examples that follow will provide a more detailed

explanation of the invention under review.

Example 1

2 g of β -cyclodextrin is added to 20 ml of an ethanol aqueous solution. This mixture is mixed with a stirrer, and as that is occurring, 100 g of EDA is added. The substance is then incubated for two hours at 50°C. After the matter has cooled at room temperature (approximately one hour), it will be mixed again, and as that mixing is occurring, it will be incubated for ten hours at 4°C. The substance that is produced is recovered by centrifugation. After it has been rinsed in n-hexane, it will be freeze dried. As a result, 1.8 g of a substance that has been taken into cyclodextrin containing 5% EDA will be produced. 1 g of this powder will then be mixed into 1 kg of a powdered milk. As a result, a homogenized milk containing a EDA content will be produced.

Example 2

The same procedures that were used in Example 1 were repeated with DGLA, ARA, and EPA. As a result of each processing, a homogenized milk containing a DGLA content, a homogenized milk containing a ARA content, and a homogenized milk containing a EPA content were produced.

Example 3

The same procedures that were used in Example 1 were

repeated with ethyl esters of EDA, DGLA, ARA, and EPA. As a result, a homogenized milk containing a EDA ethyl ester content, a homogenized milk containing a DGLA ethyl ester content, a homogenized milk containing a ARA ethyl ester content, and a homogenized milk containing an EPA ethyl ester content were produced.

Example 4

20 g of an oil-and-fat bacteria, which was produced by a culture of *Mortierella-Aeromonas* SA M0219 (FERM P-8703), was esterified using an anhydride ethanol hydrochloric acid in processing lasting over a period of three hours at 50°C. The matter was then extracted in n-hexane to produce 15 g of a fatty acid ester. The composition of this substance was 16% palmitic acid ethyl, 5% stearic acid ethyl, 27% oleic acid ethyl, 10% linoleic acid ethyl, 4% GLA ethyl, 1% EDA ethyl, 7% DGLA ethyl, 20% ARA ethyl, and 10% EPA ethyl. In the same procedures as those employed in Example 1, 2 g of this fatty acid ethyl ester compound was put to use. As a result, a homogenized powdered milk was produced. In addition, when 0.1 g of the substance taken into cyclodextrin was mixed into 1 l of a liquid milk, a homogenized liquid milk was produced.

Example 5

5 ml of a 50% ethanol aqueous solution was added to 20 g of β -cyclodextrin. The substance was added to milk that had been

kept at 60°C. 2 g of DGLA ethyl was added to it, and the matter was mixed slowly for three hours. After cooling at room temperature (for two hours), the matter was incubated for ten hours at 4°C. 1 l of water was then added, and the matter was incubated for one hour. 1 l of water was added, and the matter was mixed for one hour. Thereafter, the deposit was recovered by centrifugation. The matter was then rinsed with n-hexane, and, after that, it was freeze dried. As a result, 8.5 g of a substance that had been taken into cyclodextrin containing 10% DGLA was produced. 2 g of this powder was mixed with 2 kg of a powdered milk, and 2 g of this powder was mixed with 150 l of a liquid milk. Both produced homogenized milks.

Example 6

The same procedures as those used in Example 5 were used with 2 g of fatty acid ethyl compounds into which GLA ethyl, EDA ethyl, DGLA ethyl, ARA ethyl, and EPA ethyl had been mixed in weight percentages of 2 : 1 : 6 : 4 : 8. Homogenized powdered milks and homogenized liquid milks were produced.

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